Original Article

Increased Renal Arterial Resistance Predicts the Course of Renal Function in Type 2 Diabetes With Microalbuminuria

Romano Nosadini,[1] Mario Velussi,[2] Enrico Brocco,[3] Cataldo Abaterusso,[4] Andrea Carraro,[5] Francesco Piarulli,[6] Giuseppe Morgia,[7] Andrea Satta,[8] Rossana Faedda,[8] Avinash Abhyankar,[9] Holgher Luthman,[9] and Giancarlo Tonolo[10]

Type 2 diabetic patients often die because of end-stage renal failure, but no definitive reliable factor predicting long-term renal outcome has been identified. We tested whether a renal arterial resistance index (R/I) ≥80, using Doppler ultrasound technique, was predictive of worsening renal function. The primary end points of the study were *1*) the course of glomerular filtration rate (GFR) and *2*) the albumin excretion rate in 157 microalbuminuric, hypertensive, type 2 diabetic patients after a 7.8-year follow-up period (range 7.1–9.2). Kaplan-Meier curves for the primary end point (decrease of GFR ≥ −3.0 ml/min per 1.73 m² per year) was two to three times more frequently observed in patients with R/I ≥80. Four- to fivefold fewer patients showed a regression to normoalbuminuria during the follow-up period from baseline microalbuminuria in the cohort with R/I ≥80. Overt proteinuria did develop in 24% of patients with R/I ≥80 and in 5% of patients with R/I <80 ($P < 0.01$). In conclusion, intrarenal arterial resistance appears to play a nontrivial role in deteriorating renal function in type 2 diabetic patients. R/I is a noninvasive diagnostic procedure, which strongly predicts the outcome of renal function in type 2 diabetic patients, even when GFR patterns are still normal. *Diabetes* 55:234–238, 2006

Diabetic nephropathy accounts for end-stage renal disease (ESRD) in ~40% of patients who need dialysis or renal transplantation in the U.S. (1) and in ~15–20% of patients in Italy. ESRD is not the mandatory fate of diabetic patients with altered albumin excretion rate (AER). In fact, it has been shown that the course of renal function is ominous in some type 2 diabetic patients, who have been called "progressors," whereas other patients show steady patterns of renal function for years or even a regression of AER from microalbuminuria to normoalbuminuria ("nonprogressors") (2–4). Similar findings have also been reported in type 1 diabetic patients (5). Extensive evidence clearly demonstrates that prolonged hyperglycemia underlines chronic complications of diabetes (6). Moreover, it has been convincingly reported that tight blood pressure control, particularly using drugs that inhibit the renin angiotensin system, significantly delay the progression of renal damage in diabetic patients with incipient or advanced nephropathy (7–10). Nevertheless, 15–20% of the diabetic patients still relentlessly develop ESRD, despite aggressive antihypertensive and hypoglycemic therapy (4,6,10,11).

Although captopril renography was once the noninvasive diagnostic method of choice for patients with renal stenosis, it has now been relegated to use in secondary screening, because the quality of other noninvasive imaging methods is highly reliable and less cumbersome (12,13). It has recently been found that a renal arterial resistance index (R/I) of ≥80, using Doppler ultrasound technique, predicts worsening renal function in patients with renal diseases other than renal artery stenosis (14,15). The aim of the present study was to investigate the capacity of R/I of predicting the course of renal function in hypertensive, type 2 diabetic patients with microalbuminuria and the clinical features of progressor patients.

RESEARCH DESIGN AND METHODS

The criteria we have used since 1997, 1998, and 1999 to recruit a group of type 2 diabetic patients are as follows: age between 35 and 65 years, onset of diabetes after the age of 35 years, and no insulin need in the first 3 years after diagnosis. All of the subjects were hypertensive (>130/85 mmHg), had microalbuminuria, and were randomly treated by ACE inhibitors (one with 10 mg/day ramipril, the other one with 40 mg/day lisinopril, and so on). Twelve to 5 mg/day thiazides and 100 mg/day atenolol in the last 3–4 years, previous to the entry in the study, and 1,500 mg/day metformin with three insulin analog, before meals, or once daily ultralente insulin injection was given to each patient. These two ACE drugs were chosen because the National Health Agency in Italy decided between 1995 and 2000 that these two drugs (with these daily dosages) were the only ones useful to prevent the onset of overt kidney failure in diabetic patients when the study started. Thus we adopted their advice, and we provided these drugs at no cost to the patients. Patients were consecutively admitted to the study and randomized blindly to either ramipril or to lisinopril treatment. During the 15-day withdrawal from antihypertensive therapy, the patients were instructed with the help of the National Centers for the Care of Diabetes Mellitus and of the General Practitioners, strictly in touch with National Centers for the Care of Diabetes Mellitus in

each center to monitor blood pressure levels. Whenever blood pressure levels rose to >165/105 mmHg, the patients received calcium channel blockers sublingually to reduce the blood pressure levels to <140/90 mmHg. Calcium blockers were chosen because these drugs were able to better control blood pressure levels without substantially modifying AER, at least according to available literature. The ethical committee approved this approach to the therapy of the patients as the only one capable to measure AER reliably, without exposing the patient to vascular risks. The administration of calcium channel blockers was used only on 12 occasions during the 8-year follow-up period in 157 patients. No adverse reaction was observed. The criterion for the administration of statins was the presence of plasma LDL cholesterol level >110 mg/dl and triglyceride >200 mg/dl. This criterion was adopted to avoid an excessive drop-out of the patients because of low compliance to drug therapy during such a long period of follow-up in a relatively large population who had already accepted a rather compelling and cumbersome series of examinations. However, the number of patients in each group was too small to draw any conclusion in this study from the hypolipidemic point of view, and nevertheless, the number of patients was not significantly different in the groups with different R/I values. Twenty-five percent of the patients were treated with 40 mg/day simvastatin and 23% with 30 g/day cholestyramine without differences between groups with R/I ≥80 or <80. The patients were randomly allotted to each of the two treatments. All of the patients received 75 mg/day aspirin. No gastroenteric bleeding or muscular rabdomyolis or acute circulating increase of transaminases was observed during the follow-up period. The definition of microalbuminuria was a median between 30 and 300 μg/mg albumin over creatinine ratio (AER) in three consecutive urine specimens collected on the spot in the outpatient diabetes clinic, in the absence of hematuria. Albumin was measured by an immunoturbidimetric technique as previously described after a 15-day withdrawal from antihypertensive therapy (4); urine creatinine was measured by a modified reaction rate kinetic technique, which overcomes the drawback of the measurement of pseudo-creatinine beside creatinine (16). With regard to coronary artery disease, we used three parameters: clinical history accompanied by electrocardiographic and ultrasound imaging changes of heart wall contraction movements. Patients had serum creatinine concentrations <1.5 mg/dl. When available in the clinical history, circulating levels of troponin were used to ascertain previous episodes of myocardial infarction. Peripheral and carotid artery disease was mainly assessed by clinical evaluation (evidence or reports of claudicatio intermittens, stroke, or transitory ischemic attacks) and ultrasound techniques to ascertain the presence of atherosclerotic plaques. A major effort was put into the study to educate the patients to stop smoking, to increase exercise, and to achieve body weight reduction, using the guidelines treatments of the U.K. Prospective Diabetes Study (UKPDS) (17) and Steno Hospital (18). The values reported in the present study, with regard to body weight, smoking habits, exercise, and HbA_{1c} (A1C) levels, indicate that we, at least partially, succeeded in achieving our goal in 70–75% of our patients and on average in the overall population. With regard to exercise, each subject did walk at a pace of 2.5–3.0 km/h at least 1 h/day. The total number of smokers was ~25% in the overall population, a percentage that is comparable or even lower than in other reports in diabetic patients. The body weight is lower than that reported by other studies in the same kind of patients, as are the HbA_{1c} patterns. We did not fully accomplish the targets indicated by the American Diabetes Association and European Association for the Study of Diabetes guidelines for the control of diabetes in all of the subjects. However, we wonder whether these targets are at the moment fully realistic in Western countries, even taking into consideration that a free health organization particularly dedicated to the care of diabetes is already operating in Italy.

The main criterion for the inclusion into the study was that a decrease of glomerular filtration rate (GFR) >1 ml/min per 1.73 m² per year had to be observed during a follow-up period between 1996 and 1997 (calculated using at least three GFR values). The rationale for this latter criterion of inclusion is that a decrease of ~1 ml/year GFR is usually observed in all Caucasian subjects aged >50–55 years, even if they are not diabetic or hypertensive (19). Thus we considered progressors to be those with a threefold increase of GFR decay above the physiological functional decline of 1 ml/min per 1.73 m² per year. We recruited 171 patients, who were divided since 1997, 1998, and 1999, into two groups: with R/I (the percentage reduction of the end-diastolic flow compared with the systolic flow, measured by Doppler ultrasonography) ≥80 or with <80, calculated as average value in the two kidneys with the difference between the two kidneys not >25%. We asked the patients to perform the measurement of GFR every year during a 7.8-year follow-up period. The baseline GFR has to be >75 ml/min per 1.73 m². Two patients developed renal cancer. Three patients developed myocardial infarction, from which one died. This event did occur not during or after at least 2–3 months after the withdrawal from antihypertensive therapy. One patient showed a three- to fourfold increase of transaminases above baseline levels. Two patients had stroke, from which one died. Six patients were excluded because they have a

difference between the R/I calculated in the two different kidneys >25%. Two of the latter six patients had untreated renal artery stenosis, resulting in a 50% reduction in the luminal diameter. All of these patients were excluded from the study. Thus 157 patients ended the study after a mean follow-up period of 7.8 years (range 7.1–9.2). Fifty-nine patients had an ultrasound Doppler R/I average index in the two kidneys ≥80 and 98 patients <80.

End points. *1*) The primary end point of the study was the evaluation of the number of patients with a decrease of GFR >3 ml/min per 1.73 m² per year during a follow-up period of 7.8 years (range 7.1–9.2). *2*) The secondary end point was the evaluation of the course of AER, expressed both as absolute values and percent change from microalbuminuria to normoalbuminuria and from microalbuminuria to proteinuria.

GFR. GFR was measured evaluating the clearance rate of ⁵¹Cr-labeled EDTA. More particularly, we evaluated GFR from the clearance rate of ⁵¹Cr-EDTA technique. At 9:00 A.M., a bolus injection of 1 μg Ci/kg ⁵¹Cr-EDTA was given over 30 s. Seven samples were collected at 5, 12.5, 50, 80, 180, 240, and 300 min, with time 0 being the midpoint of the infusion interval. GFR was measured eight times: at baseline and every year from the 2nd until the 8th year of the follow-up period. The coefficient of variation of the bolus injection, because of measurement errors related to the calculation of the radioactivity concentration of the dose injected of each sample using a 40-min time for count measurement in duplicate, was 2.6 ± 0.3%. The coefficient of variation of repeated measurements, which accounts for physiological variability in different days in the same subject, was 3.5 ± 0.8%. Further details have been provided previously (20,21). GFR courses of individuals were modeled using an adaptation of smoothing and regression cubic B-splines. Group comparisons were based on five component vectors of fitted GFR values using a permutation approach to a Hotelling's T^2 statistic (22). A similar, although not identical, approach had been previously used by Nosadini et al. (4). Blood samples were collected after an overnight fast for lipid measurement. Serum triglycerides and total cholesterol were measured by enzymatic techniques using commercially available Konelab 20 Autoanalyzer (Thermo Clinical Labsystems, Vantaa, Finland).

Ultrasound Determination of the R/I. A Sienna Sonoline ultrasound machine (Siemens, Milan, Italy) with a 3.5-MHz convex-array transducer was used. The B-mode measurements were performed at the same time as the Doppler measurement of the R/I. The ultrasonographic procedure that we adopted has been described previously (14,15,23,24). Briefly, the maximal length, width, and depth of the kidney were determined, and the renal volume was calculated as one-half the product of the three dimensions. The renal parenchimal width was measured from the capsulated to the tip of a renal pyramid in both kidneys. Intrarenal Doppler signals were obtained from two to three representative proximal segmental arteries (the first vessels branching of the main renal artery) from both kidneys. The peak systolic velocity (V_{max}) and the minimal diastolic velocity (V_{min}) were determined, the R/I was calculated as $100 \times [1 - (V_{min}:V_{max})]$, and the results from the two or three measurements were averaged. The reproducibility of R/I index measurements was tested in 45 patients by two independent investigators in 3 days in 3 consecutive weeks to calculate the intraobserver-intrasession variability, the intraobserver-intersession variability, and the interobserver-intrasession variability. The respective values for coefficient of variation were 1.7, 3.3, and 2.7%. To rule out renal-artery stenosis, the course of the renal artery was determined with color-flow imaging. Stenosis was suspected if a segmental of the vessel showed color-flow abnormality. The maximal systolic-flow velocity was measured at the site of "aliasing" (disturbance) (V_{sten}) and the point most distal to the site (V_{postem}). The area of stenosis (as percentage of the total area) was calculated according to the continuity equation as $100 \times [1 - (V_{postem}:V_{sten})]$. According to this technique, an area of stenosis >75%, which approximately results in a reduction in the diameter of the vessel >50%, was diagnosed only when V_{sten} was higher than V_{postem} by a factor >4.

Percutaneous kidney biopsies. Percutaneous kidney biopsies were performed under ultrasound guidance in the patients at baseline. Tissue was examined under a dissecting microscope to make sure of an adequate number of glomeruli in the kidney specimen. A 1-mm core was immediately snap frozen in liquid nitrogen for immunofluorescence. A second fraction was placed in Zenker's fixative embedded in paraffin and processed for light microscopy. The patterns of renal lesions in the 157 type 2 diabetic patients with hypertension and microalbuminuria were classified as described previously using light microscopy technique by A.S. and R.F. (4,11,25,26), who were unaware of the clinical features of the patients as follows:

Category 1. Sixty-six patients had typical diabetic nephropathology. These biopsies showed established diabetic glomerulopathy with proportionally severe tubulointerstitial and arteriolar changes. This picture is typical of that seen in type 1 diabetic patients. These patients had proliferative diabetic retinopathy.

Category 2. Forty patients had atypical patterns of renal injury. These patients had absent or only mild glomerular diabetic changes with dispropor-

TABLE 1
Demographic and clinical characteristics of the hypertensive, microalbuminuric type 2 diabetic patients at baseline

| Characteristics | R/I | | P value |
	≥80	<80	
n	59	98	
Age (years)	62 ± 8	64 ± 9	NS
Duration of diabetes (years)	13 ± 4	12 ± 5	NS
Systolic/diastolic blood pressure (mmHg)	132/80 ± 11/5	133/79 ± 13/7	NS
Plasma cholesterol (mg/dl)	221 ± 15	189 ± 16	$P < 0.05$
Plasma triglycerides (mg/dl)	187 ± 32	147 ± 31	$P < 0.05$
A1C (%)	7.3 ± 0.6	7.4 ± 0.7	NS
Sex (male/female)	40/19	60/38	NS
Coronary artery disease (%)	41	12	$P < 0.01$
Carotid artery disease (%)	31	9	$P < 0.01$
Peripheral artery disease (%)	40	19	$P < 0.01$
Renal histological category 1 (n)	36/59	30/98	$P < 0.01$
Renal histological category 2 (n)	19/59	21/98	$P < 0.01$
Renal histological category 3 (n)	4/59	47/98	$P < 0.01$
Smoking habit (%)	39	12	$P < 0.01$

Data are means ± SD. Blood pressure and glucose control values were those during the entire period of follow-up with drug treatments.

tionately severe renal structural lesions, including tubulointerstitial lesions, advanced glomerular arteriolar hyalinosis, and/or global glomerular sclerosis (>25%) in the presence of absence or mild mesangial expansion (index of mesangial expansion <0.5%). These patients had background diabetic retinopathy (60%), whereas 40% had no diabetic retinopathy.

Category 3. Fifty-one patients had normal renal structure. These biopsies were normal or showed very mild mesangial expansion, tubulointerstitial changes, or arteriolar hyalinosis in any combination. These patients did not have proliferative diabetic retinopathy (39% background retinopathy and 61% no diabetic retinopathy).

The analysis of kidney specimens was carried out according to the score system described by Mauer et al. (25) and Fioretto et al. (26). The study was approved by the ethical committee of the University of Padova and Sassari.

Statistical analysis. Data are reported as means ± SE or SD when normally distributed and as medians with ranges when the data were not normally distributed. The unpaired and paired Student's t test for parametric parameters, the Wilcoxon test for nonparametric parameters, and the quadratic χ^2 test for the comparison of the data expressed as percentages were evaluated using the software SPSS analysis approach. Because the number of subjects in each of the two groups of comparison was uneven, we used the formula of Fleiss et al. (27) to calculate the size of the cohort of patients we needed, assuming a difference between groups with regard to the primary end point of $1 - P = 0.4$, with P being the relative frequency of the exposed controls and an odds ratio statistically significant at $P < 0.05$ with a size dimension of the sample >80%. The number of patients in the group with R/I ≥80 was 59, and the number in the group with R/I <80 was 98 (27,28). Kaplan-Meier analysis with log-rank test was used as appropriate to assess the differences between groups, after correction for metabolic and systemic parameters. The number of years of follow-up was calculated from the date of ultrasonography until the date of the last documented visit in the out patient clinic, encompassing the GFR measurement (on average after 7.8 years). For multivariate analysis, the effect of multiple variables on worsening of renal function was evaluated using stepwise forward Cox regression analysis (with $P < 0.10$ as threshold level of significance for the removal of the variable from the analysis or the definition of nonsignificativity and $P < 0.05$ as the threshold for entry into the model). The variables investigated were the R/I; age; duration of the disease; sex; clinical, biochemical, electrocardiographic, or ultrasound signs of atherosclerosis in the heart, legs, or central nervous system; AER; average systolic, diastolic, and A1C patterns during the 7.8-year follow-up period; and baseline GFR and lipid circulating values.

RESULTS

The clinical characteristics of the two groups of type 2 hypertensive, microalbuminuric, diabetic patients with R/I at baseline ≥80 and <80 are shown in Table 1. Patients with R/I ≥80 showed slightly higher circulating lipid patterns and stronger smoking habits. Blood pressure control was reasonably tight, with average levels of systolic and diastolic blood pressure <140/90 mmHg even if

not <130 mmHg as for systolic levels, as recommended by the guidelines for the treatment of hypertension in diabetes. BMI was 27 ± 0.4 and 28 ± 0.6 kg/m² at baseline, respectively, in the groups. Patients with R/I ≥80 more frequently showed clinical, biochemical, and ultrasound evidence of atherosclerosis (41 vs. 12% coronary artery disease, 31 vs. 9% carotid artery disease, and 40 vs. 19% peripheral artery disease, Table 1). Patients with R/I ≥80 more frequently had renal histological patterns of category 1 and 2 than 3 (category 1, 61 vs. 30%; category 2, 32 vs. 21%; and category 3, 7 vs. 48%; see absolute number in Table 1). The number of patients who smoked was greater among patients with R/I ≥80 than with R/I <80. Kaplan-Meier curves for the primary end point (decrease of GFR ≥ −3.0 ml/min per 1.73 m² per year), corrected as for lipid, A1C patterns, blood pressure levels, renal histology category, and smoking habits, was two to three times more frequently observed in patients with R/I ≥80 (Fig. 1). The percentage of patients who showed a regression to normoalbuminuria during the follow-up period was four- to fivefold lower in patients with R/I ≥80 than in those with R/I <80 (Table 2). On the contrary, overt proteinuria did develop more frequently in patients with R/I ≥80 than in patients with R/I <80 (24 vs. 5%, $P < 0.01$). Average AER levels decreased significantly in patients with R/I <80 but not in those with R/I ≥80 (Table 2). Multivariate analysis revealed a number of variables that differed significantly between patients with R/I ≥80 or with R/I <80 with regard to the primary end point (decrease of GFR ≥ −3.0 ml/min per 1.73 m² per year). The strongest risk factor was the R/I index (Table 3). Also, the presence of combined artery disease, renal histological category, and smoking habits played a discriminatory role, which was, however, not so important in comparison with the R/I index, as evidenced by the lower relative risks associated with these variables (Table 3).

The size of the cohort of patients with a R/I value ≥80 or <80 with systolic blood pressure >140 mmHg was too low to allow statistical comparisons. The cohort of patients, within the entire population, who showed an average value of systolic blood pressure >140 mmHg (19%) had a greater decline of GFR than those in whom systolic blood pressure was steadily maintained <140 mmHg (−2.56 ±
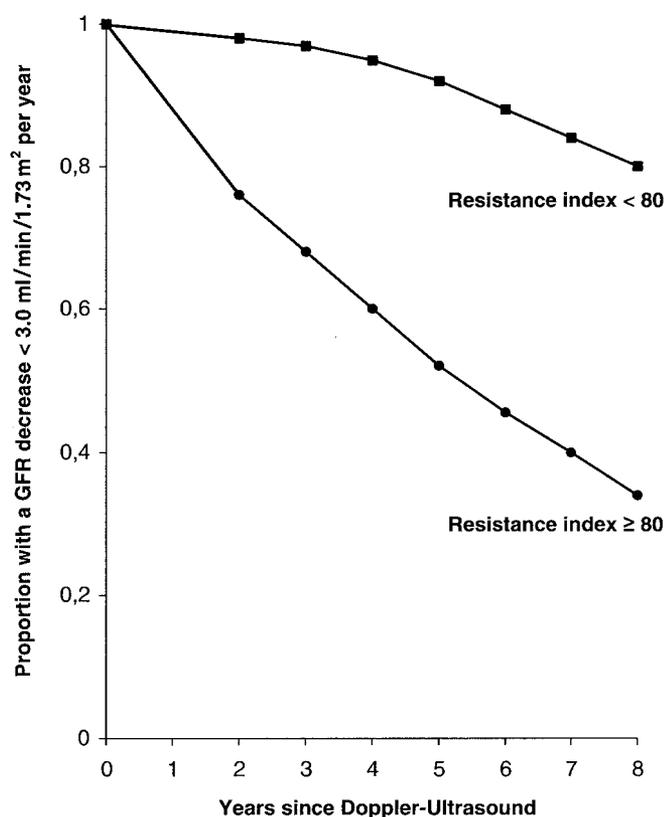
FIG. 1. Kaplan-Meier analysis, corrected for lipid, A1C, blood pressure levels, and smoking habits, of time to the predefined primary end point, e.g., an yearly decline of GFR ≥−3 ml/min per 1.73 m² in the 59 patients with an R/I index ≥80 and in the 98 patients with an R/I index <80 during the 7.8-year follow-up period. The percentage of patients with a yearly decrease of GFR ≥−3 ml/min per 1.73 m² was two to three times greater in patients with an R/I ≥80.

0.111 vs. -1.23 ± 0.12 ml/min per 1.73 m² per year, $P < 0.05$). The number of patients with steady values of diastolic blood pressure >90 mmHg was too small to allow any comparison. The number of patients treated by simvastatin was too low inside each group of patients with different R/I to allow a statistical comparison with those treated with different hypocholesterolemic drugs or untreated. Therefore we just compared the rate of decay of GFR between patients treated by simvastatin or untreated within the entire population. We observed a slightly lower rate of decay in the patients using simvastatin than in the other patients (-1.96 ± 0.29 vs. -2.77 ± 0.37, $P < 0.07$, NS).

DISCUSSION

Microalbuminuria is not merely a predictor of diabetic nephropathy but also constitutes evidence of renal dam-

TABLE 3
Relative risk (RR) of a decrease of GFR per year >3.0 ml · min⁻¹ · 0.1.73 m⁻² according to selected variables (multivariate analysis) with relative P values

Variable	Relative risk	P value
R/I ≥80 at baseline	10.7 (3.3–15.1)	$P < 0.0001$
Baseline GFR	1.8 (0.8–2.9)	NS
Baseline AER	1.9 (0.7–3.2)	NS
Combined artery disease	5.2 (1.3–7.1)	$P < 0.05$
Renal histological category at base line	6.1 (2.0–12.4)	$P < 0.01$
Duration of the disease	1.1 (0.6–1.8)	NS
A1C during follow-up	1.2 (0.4–2.2)	NS
Blood pressure during follow-up	1.3 (0.6–2.7)	NS
Smoking habit (%)	4.3 (1.1–5.9)	$P < 0.05$

Data are means (95% CI).

age. This has been demonstrated by Adler et al. (29) and by our research group (11), who showed in microalbuminuria altered mRNA expression of several glomerular proteins.

Patterns of glycemic control accomplishing an average A1C level of 7.0% are able to delay the onset of overt diabetic nephropathy in type 2 diabetic patients (17). We achieved an average value of A1C of 7.3–7.4%, despite thorough efforts to obtain a tighter glycemic control. Nevertheless even in the UKPDS study (17), a nontrivial fraction of the patients had A1C levels >7.0%, and several other reports in type 2 diabetic patients with altered AER showed A1C levels close to 8.0% (7–11).

The R/I index is significantly correlated with increased blood pressure and decreased renal function (14,15,23,30). We suggest that R/I index ≥80 can predict the outcome of renal function in type 2 diabetic patients with microalbuminuria. This finding might be important from three different points of view. First, ischemic atherosclerotic injury, at the level of renal small vessels, might accelerate the decay of GFR, possibly further deteriorating the molecular structural characteristics of endothelial-podocyte system. Second, R/I index can be used to predict the outcome of renal function in type 2 diabetic patients with microalbuminuria. Third, an increased R/I index is closely associated, in type 2 diabetic patients with microalbuminuria, with patterns of glomerular lesions of typical or atypical diabetic nephropathy. The latter observation resembles the close relationship between proliferative diabetic retinopathy and patterns of glomerular and renal lesions typical of diabetic nephropathy that we have described previously (4,11,26). However, diabetic retinopathy appears to be a less specific marker of the progression of renal damage, because a nontrivial fraction of the patients who have nonproliferative background retinopa-

TABLE 2
GFR and AER at baseline and after 8 years in patients with R/I ≥80 or <80 and patients showing regression to normoalbuminuria and progression to proteinuria

	Group 1 (R/I <80)		Group 2 (R/I ≥80)	
	Baseline	8 years	Baseline	8 years
GFR (ml · min⁻¹ · 1.73 m⁻²)	98 ± 2	91 ± 7	97 ± 11	70 ± 8*†
AER (μg/mg)	81 (32–240)	40 (11–339)*	88 (31–261)	81 (13–570)
Normoalbuminuria (%)	—	31	—	7†
Proteinuria (%)	—	5	—	24†

Data are means ± SE, median (range), and %. *$P < 0.05$ baseline vs. 8 years. †$P < 0.01$ group 1 vs. group 2.

thy may not develop overt nephropathy (4). In fact, only type 2 diabetic patients with proliferative retinopathy are bound to show a rapid decline of GFR, but only 10–20% of type 2 diabetic patients have proliferative retinopathy (4,11,26). Moreover, the ophthalmoscopic examination, although mandatory to prevent and to treat diabetic proliferative retinopathy, is not less cumbersome than the evaluation of R/I to prevent and predict diabetic nephropathy. With regard to vascular events, the occurrences of carotid, peripheral, and coronary pathology are also less specific to predict the development of overt nephropathy than R/I index, because they may frequently be found also in type 2 diabetic patients without microalbuminuria and predisposition to develop overt nephropathy.

Thus we suggest that the R/I index might be useful to identify the cohort of microalbuminuric patients with more severe renal lesions and those prone to develop a rapid decay of GFR without performing routinely the invasive procedure of renal biopsy. Krolewski et al. (31,32) and Viberti et al. (33) demonstrated some years ago that a cohort of type 1 diabetic patients have a predisposition to develop overt diabetic nephropathy in association with increased Na/Li countertransport activity in erythrocytes, a marker of arterial hypertension. We have confirmed this observation in Caucasian type 2 diabetic patients (34). Arterial renal resistance, which also is frequently associated with arterial hypertension (30) and might reflect a more severe endothelial dysfunction, appears to be a further feature of this cohort of patients prone to develop overt nephropathy. Smoking habits certainly contributed to explain the rapid decline of GFR in our patients, but the role of R/I index emerged as the predominant risk factor (Table 3). Clearly, our findings need to be confirmed by other authors both in Caucasian and non-Caucasian type 2 diabetic patients.

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